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CLAIM CONVENTION PRIORITY

SIRS:

Applicant hereby claims benefit of Convention Priority based

(1) Japan SN 1999-149399

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Filed herewith are the English translation thereof together with verification by the translator.

Accordingly, applicant being beneficiary ofConvention Priority is hereby should be awarded Convention Priority.

Respectfully

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DECLARATION

I, Toshiyuki Ueno, a member of Yokogawa Electric Corporation (Yokogawa Denki Kabushiki Gaisha) having a principal place of business at 9-32, Nakacho 2-chome, Musashino-shi, Tokyo 180-8750 Japan, do solemnly declare that the attached documents are full, true and faithful translation made by me this 30th day of June 2008 of a certified copy of the Japanese Patent Application No. 2000-007724 "Biochip Reader" consisting of Application for certificate dully certified thereon and Specification. And I make this solemn declaration conscientiously believing the same to be true.

T. Ueno

Toshiyuki Ueno

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[Name of Article] Abstract 1

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Date of Submission: January 17, 2000

Reference Number = N990115 Tokuganhei 2000-007724

[Document Name] Specification

[Title of the Invention] Biochip Reader

[What Is Claimed Is]

[Claim 1]

A biochip reader comprising:

a light source for emitting excitation light;

a dichroic mirror for reflecting said excitation light or allowing said excitation light to pass through said dichroic mirror;

an objective lens for condensing light that has been reflected by or passed through said dichroic mirror onto a biochip and projecting fluorescent light produced at said biochip onto said dichroic mirror;

an optical detector for detecting said fluorescent light; and

a lens for condensing said fluorescent light that has been reflected by or passed through said dichroic mirror onto said optical detector,

wherein said biochip is configured using a transparent substrate allowing for passage of said excitation light and said fluorescent light and said excitation light is irradiated from one side of said biochip opposite to the side where samples are arranged.

[Claim 2]

A biochip reader as defined in claim 1, wherein said objective lens is an immersion lens.

[Claim 3]

A biochip reader as defined in claim 1, wherein said objective lens is a water immersion lens or an oil immersion lens.

[Claim 4]

A biochip reader as defined in claim 1, wherein said objective lens is an SIL.

[Claim 5]

A biochip reader as defined in claim 1, wherein an optical system is a confocal optical system.

[Claim 6]

A biochip reader as defined in claim 1, wherein an anti-reflection coating is formed on one side of said biochip opposite to the side where samples are arranged.

[Claim 7]

A biochip reader as defined in claim 1, wherein transparent electrodes are formed on a surface of said transparent substrate.

[Claim 8]

A biochip reader as defined in claim 7, wherein said transparent electrodes are made of an indium-tin oxide film.

[Claim 9]

A biochip reader as defined in claim 1, wherein said samples are DNA segments.

[Claim 10]

A biochip reader as defined in claim 1, wherein said samples are RNA segments.

[Claim 11]

A biochip reader as defined in claim 1, wherein said samples are protein segments.

[Claim 12]

A biochip reader as defined in claim 1, wherein said samples are sugar chain segments.

[Detailed Description of the Invention]

[0001]

[Technical Field to Which the Invention Pertains]

The present invention relates to a reader for biochips, such as DNA

chips and protein chips. More specifically, the invention relates to a reader whose S/N ratio is superior and whose cost can be reduced.

[0002]

[Description of the Prior Art]

A biochip, such as a DNA chip, used with the reader has the structure in which several thousand to several ten thousand types of known DNA segments are arranged in arrays on a substrate. If any unknown DNA segment is flowed onto the DNA chip, it combines with a DNA segment of the same type. Taking advantage of this nature of DNA, a known DNA segment that has formed a combination is examined by the biochip reader to identify the properties of the unknown DNA, such as DNA arrangement.

[0003]

FIG. 5 shows an example of hybridizing such a biochip as described above. In FIG. 5, the six types of DNA segments, DN01 to DN06, are arranged in arrays on a substrate SB01 to form a DNA chip.

[0004]

UN01 is an unknown DNA segment and is previously given a fluorescent mark, as indicated by LM01 in the figure. When hybridized to the DNA chip, this unknown DNA segment combines with another DNA segment whose arrangement is complementary.

[0005]

For example, the unknown DNA segment UN01 in FIG. 5 combines with the known DNA segment DN01, as indicated by CB01 in FIG. 5.

[0006]

Using the biochip reader, excitation light is irradiated at the DNA chip thus hybridized, in order to detect fluorescent light emitted from the fluorescent mark described earlier. Consequently, it is possible to know which of the known DNA segments the unknown DNA segment has combined with.

[0007]

For example, in an image resulting from scanning the DNA chip indicated by SI01 in FIG. 5, fluorescent light is observed only at a spot where the DNA combination CB01 has been produced. This means fluorescent light is detected only from the spot indicated by LD01 in FIG. 5.

[0008]

FIG. 6 is a schematic block diagram showing an example of the conventional biochip reader described earlier. In FIG. 6, the numeral 1 indicates a light source for emitting excitation light, such as a laser light source, the numeral 2 indicates a dichroic mirror, the numeral 3 indicates an objective lens, the numeral 4 indicates a DNA chip which is a biochip onto which multiple cells are arranged in arrays, the numeral 5 indicates a filter, the numeral 6 indicates a lens, and the numeral 7 indicates an optical detector, such as a photomultiplier tube.

[0009]

The symbols CL01 to CL03 are the aforementioned cells in which DNA segments, namely samples, of the same type are arranged.

[0010]

Light emitted from the light source 1 is reflected by the dichroic mirror 2 as excitation light and condensed onto cells on the DNA chip 4 through the objective lens 3. For example, the excitation light is

condensed onto the cell CL02 in FIG. 6.

[0011]

Fluorescent light produced by the excitation light in the cell CL02 becomes parallel light as it travels through the objective lens 3, and passes through the dichroic mirror 2. Fluorescent light that has passed through the dichroic mirror 2 travels through the filter 5 and is condensed onto the optical detector 7 by the lens 6.

[0012]

The DNA chip 4 is scanned by a drive means which is not shown in FIG. 6. For example, the DNA chip 4 is scanned in the direction indicated by MV01 in FIG. 6 so that the excitation light is irradiated at the remaining cells CL01 and CL03 on the DNA chip 4.

[0013]

Consequently, it is possible to identify the arrangement of the unknown DNA segment from the position of a cell where fluorescence has taken place.

[0014]

[Problems to Be Solved by the Invention]

Dust may deposit on the DNA chip 4, however, for such reasons as the mixing of foreign matter with a liquid in which the unknown DNA segment is hybridized or the way subsequent processes are carried out. If the dust is organic, the excitation light causes the dust to emit fluorescent light that is more intense than that emitted by a cell. This results in the problem that the fluorescent light serves as noise and therefore deteriorates the S/N ratio.

[0015]

FIG. 7 is an enlarged view of the cell CL02 shown in FIG. 6. Members indicated by 3, 4 and CL02 are the same as those in FIG. 6. If the DNA chip 4 is contaminated with dust particles marked DS01 and DS02 in FIG. 7, fluorescent light indicated by LL01 is produced by the excitation light in addition to fluorescent light emitted from the cell CL02. This deteriorates the S/N ratio.

[0016]

For this reason, a confocal optical system has been used with the conventional biochip reader to detect only the fluorescent light produced by cells by removing fluorescent light produced by dust. Alternatively, a DNA chip has been hermetically sealed to prevent it from being contaminated with dust. However, these measures have caused the problems not only of increased cost but also of insufficiently improved S/N ratio.

The objective of the present invention is therefore to provide a biochip reader whose S/N ratio is superior and whose cost can be reduced.

[0017]

[Means for Solving Problems]

In order to achieve the aforementioned object, the present invention provides, as defined in claim 1, a biochip reader comprising:

a light source for emitting excitation light;

a dichroic mirror for reflecting the excitation light or allowing the excitation light to pass through the dichroic mirror;

an objective lens for condensing light that has been reflected by or

passed through the dichroic mirror onto a biochip and projecting fluorescent light produced at the biochip onto the dichroic mirror;

an optical detector for detecting the fluorescent light; and

a lens for condensing the fluorescent light that has been reflected by or passed through the dichroic mirror onto the optical detector,

wherein the biochip is configured using a transparent substrate allowing for passage of the excitation light and the fluorescent light and the excitation light is irradiated from one side of the biochip opposite to the side where samples are arranged, thereby improving the S/N ratio of the biochip reader and allowing for a cost reduction therein.

[0018]

According to claim 2 in the present invention, the biochip reader as defined in claim 1 comprises an immersion lens as the objective lens, thereby improving the numerical aperture and hence increasing the S/N ratio further.

[0019]

According to claim 3, the biochip reader as defined in claim 1 comprises a water immersion lens or an oil immersion lens as the objective lens, thereby improving the numerical aperture and hence increasing the S/N ratio further.

[0020]

According to claim 4, the biochip reader as defined in claim 1 comprises an SIL as the objective lens, thereby improving the numerical aperture and hence increasing the S/N ratio further.

[0021]

According to claim 5, the biochip reader as defined in claim 1 comprises a confocal optical system as the optical system thereof, thereby further increasing the S/N ratio than when a non-confocal optical system is used.

[0022]

According to claim 6, the biochip reader as defined in claim 1 has a feature wherein an anti-reflection coating is formed on one side of the biochip opposite to the side where samples are arranged, thereby increasing the luminous energy of excitation light irradiated at the samples and hence allowing for an improvement in the S/N ratio.

[0023]

According to claim 7, the biochip reader as defined in claim 1 has a feature wherein transparent electrodes are formed on a surface of the transparent substrate, thereby increasing the probability for samples having a complementary sequence to combine with each other.

[0024]

According to claim 8, the biochip reader as defined in claim 7 has a feature wherein transparent electrodes made of an indium-tin oxide film are formed on a surface of the transparent substrate, thereby increasing the probability for samples having a complementary sequence to combine with each other.

[0025]

According to claim 9, the biochip reader as defined in claim 1 has a feature wherein the samples are DNA segments and known samples having a

complementary sequence are combined by hybridization with unknown samples marked with a fluorescent substance, enabling the sequence of the unknown samples to be identified.

[0026]

According to claim 10, the biochip reader as defined in claim 1 has a feature wherein the samples are RNA segments and known samples having a complementary sequence are combined by hybridization with unknown samples marked with a fluorescent substance, enabling the sequence of the unknown samples to be identified.

[0027]

According to claim 11, the biochip reader as defined in claim 1 has a feature wherein the samples are protein segments and known samples having a complementary sequence are combined by antigen-antibody reaction with unknown samples marked with a fluorescent substance, enabling the sequence of the unknown samples to be identified.

[0028]

According to claim 12, the biochip reader as defined in claim 1 has a feature wherein the samples are sugar chain segments and known samples having a complementary sequence are combined by antigen-antibody reaction with unknown samples marked with a fluorescent substance, enabling the sequence of the unknown samples to be identified.

[0029]

[Mode for Carrying out the Invention]

The present invention is described in detail below with reference to

the accompanying drawings. FIG. 1 is a schematic block diagram showing one embodiment of a biochip reader in accordance with the present invention.

In FIG. 1, members indicated by 1 to 3 and 5 to 7 are the same as those in FIG. 6, and the numeral 8 indicates a DNA chip using a plastic or glass substrate which allows excitation light or fluorescent light to pass through it.

[0030]

Members indicated by CL11 to CL13 are cells the same as those described earlier on which multiple samples of DNA segments of the same type are arranged. The symbols DS11 and DS12 indicate dust particles adhering to the cell CL12 on the DNA chip 8.

[0031]

Light emitted as excitation light from a light source 1 is reflected by a dichroic mirror 2 and condensed onto a cell on the DNA chip 8 through an objective lens 3. At this point, the excitation light is irradiated from the side opposite to the side where the cells are arranged.

[0032]

For example, the excitation light is irradiated at the cell CL12 through the transparent substrate of the DNA chip 8.

[0033]

Fluorescent light produced by the excitation light at the cell is made parallel through the objective lens 3, and passes through the dichroic mirror 2. The fluorescent light that has passed through the dichroic mirror 2 is condensed by a lens 6 onto the optical detector 7 through a filter 5. At this point, the fluorescent light produced by the excitation

light at the cell passes through the DNA chip 8 and is output to the side opposite to the side where the cells are arranged.

[0034]

The DNA chip 8 is scanned by a drive means which is not shown in the figure. For example, the DNA chip 8 is scanned in the direction indicated by MV11 in FIG. 1 so that the excitation light is irradiated at the remaining cells CL11 and CL13 on the DNA chip 8.

[0035]

Liquid in which unknown DNA segments are hybridized is flowed onto the side where the cells, such as the cell CL12, shown in FIG. 1 are arranged. The dust particles DS11 and DS12 adhere to the side of the substrate where the cells are arranged on the DNA chip 8.

[0036]

On the other hand, no foreign matter such as the dust particle DS11 adheres to the side opposite to the side where the cells are arranged on the DNA chip 8.

[0037]

Consequently, fluorescent light resulting from the dust particle and serving as a noise component can be reduced by irradiating the excitation light from the side opposite to the side where the cells are arranged on the DNA chip 8. For example, the excitation light is irradiated neighboring a boundary between the substrate of the DNA chip 8 and a cell.

[0038]

In addition, a simple optical system can be used with the biochip reader and there is no need for hermetically sealing the DNA chip 8. These

advantages make it possible to reduce the cost of the biochip reader.

[0039]

It should be noted that although only a DNA chip is shown as an example of biochips when explaining figures, including FIG. 1, the biochips are, as a matter of course, not limited to a DNA chip only. They may be such chips as fabricated by arranging in arrays segments of ribonucleic acid (RNA), protein or sugar chain on a transparent substrate.

[0040]

In this case, RNA segments undergo hybridization as with DNA segments, while protein segments and sugar chain segments are submitted to an antigen-antibody reaction. In either case, segments of known samples combine with segments of unknown samples marked with a fluorescent substance.

[0041]

In addition, although the objective lens 3 shown in FIG. 1 is of the non-immersion type, it may be of the immersion type, such as a water immersion or an oil immersion lens. FIG. 2 is a partially enlarged view of the cell CL12 shown in FIG. 1 when an immersion lens is used. Members indicated by 3, 8 and CL12 in FIG. 2 are the same as those in FIG. 1.

[0042]

In FIG. 2, the symbol LQ11 indicates a fluid such as water or oil filled into the gap between the objective lens 3 and the DNA chip 8. In this arrangement the numerical aperture (NA) is improved, thereby improving the S/N ratio further, because of the refractive index of the fluid, such as water or oil. For this arrangement, however, the method in which beams

of excitation light itself are scanned is more suitable than scanning the DNA chip 8 or the objective lens 3.

[0043]

FIG. 3 is a partially enlarged view of the cell CL12 shown in FIG. 1 when a solid immersion lens (SIL), which has the same effect as an immersion lens, is used. In FIG. 3, members indicated by 8 and CL12 are the same as those in FIG. 1, and the numeral 9 indicates an SIL. Also, in this arrangement the numerical aperture (NA) is improved by the SIL, thereby improving the S/N ratio further.

[0044]

If the substrate of a DNA chip 8 needs to be conductive, it may be prepared by placing transparent electrodes made of an indium-tin oxide (ITO) film on a transparent substrate. Hybridization can be accelerated by applying a positive voltage to the electrodes because DNA is charged with negative electricity.

[0045]

An anti-reflection coating may be placed on the surface of the DNA chip 8's substrate opposite to the surface where cells are arranged.
FIG. 4 is a schematic view showing a comparison between DNA chips with and without an anti-reflection coating. In FIG. 4, members indicated by 8 and CL12 are the same as those in FIG. 1, and the numeral 10 indicates an anti-reflection coating.

[0046]

The structure of the DNA chip 8 shown in FIG. 4 (A) is the same as the one shown in FIG. 1. In FIG. 4 (B), the anti-reflection coating 10 is

formed on one side of the substrate of the DNA chip 8 opposite to the side where cells are arranged. In the case of FIG. 4 (A), the ratio of reflected light RL01 to incident light IL01 is approximately "4%". In the case of FIG. 4 (B), however, the ratio of reflected light RL11 to incident light IL11 can be reduced to as small as approximately "0.5%". Consequently, the luminous energy of excitation light irradiated at cells on the DNA chip 8 increases, improving the S/N ratio.

[0047]

The side of the substrate of the DNA chip 8 where cells are arranged may be in the state of dryness. It is also possible to leave that side of the substrate wetted with hybridization liquid.

[0048]

Although a laser light source has been mentioned earlier as an example of the source of excitation light, a non-laser light source, such as an LED lamp, xenon lamp, halogen lamp or any other white light source, may be used instead.

[0049]

If a confocal optical system is used with the biochip reader, fluorescent light produced by dust particles can be removed more effectively. Consequently, it is possible to improve the S/N ratio further, compared with biochip readers with a non-confocal optical system.

[0050]

[Effect of the Invention]

As described above, the following advantages are offered in accordance

with the present invention.

According to claim 1 in the present invention, the excitation light is irradiated from one side of the biochip opposite to the side where samples are arranged, thereby improving the S/N ratio and allowing for a cost reduction in the biochip reader.

[0051]

According to claims 2 to 4, an immersion lens or an SIL is used as the objective lens, thereby improving the numerical aperture (NA) and hence increasing the S/N ratio further.

[0052]

According to claim 5, the optical system is configured using a confocal optical system, thereby further increasing the S/N ratio than when a non-confocal optical system is used.

[0053]

According to claim 6, an anti-reflection coating is formed on one side of the biochip opposite to the side where samples are arranged, thereby increasing the luminous energy of excitation light irradiated at the samples and hence allowing for an improvement in the S/N ratio.

[0054]

According to claims 7 and 8, transparent electrodes are formed on a surface of the transparent substrate, thereby making it possible to accelerate hybridization by applying a positive voltage to the electrodes because DNA is charged with negative electricity.

[0055]

According to claims 9 and 10, the samples are RNA segments or protein

segments and, therefore, known samples having a complementary sequence are combined by hybridization with unknown samples marked with a fluorescent substance, enabling the sequence of the unknown samples to be identified.

[0056]

According to claims 11 and 12, the samples are protein segments or sugar chain segments and, therefore, known samples having a complementary sequence are combined by antigen-antibody reaction with unknown samples marked with a fluorescent substance, enabling the sequence of the unknown samples to be identified.

[Brief Description of the Drawings]

[FIG. 1]

FIG. 1 is a schematic block diagram showing one embodiment of a biochip reader in accordance with the present invention.

FIG. 2 is a partially enlarged view of a cell when an immersion lens is used.

FIG. 3 is a partially enlarged view of a cell when a solid immersion lens (SIL) is used.

FIG. 4 is a schematic view showing a comparison between DNA chips with and without an anti-reflection coating.

FIG. 5 is a schematic view showing an example of hybridization seen in biochips.

FIG. 6 is a schematic block diagram showing an example of a conventional biochip reader.

FIG. 7 is an enlarged view of a cell.

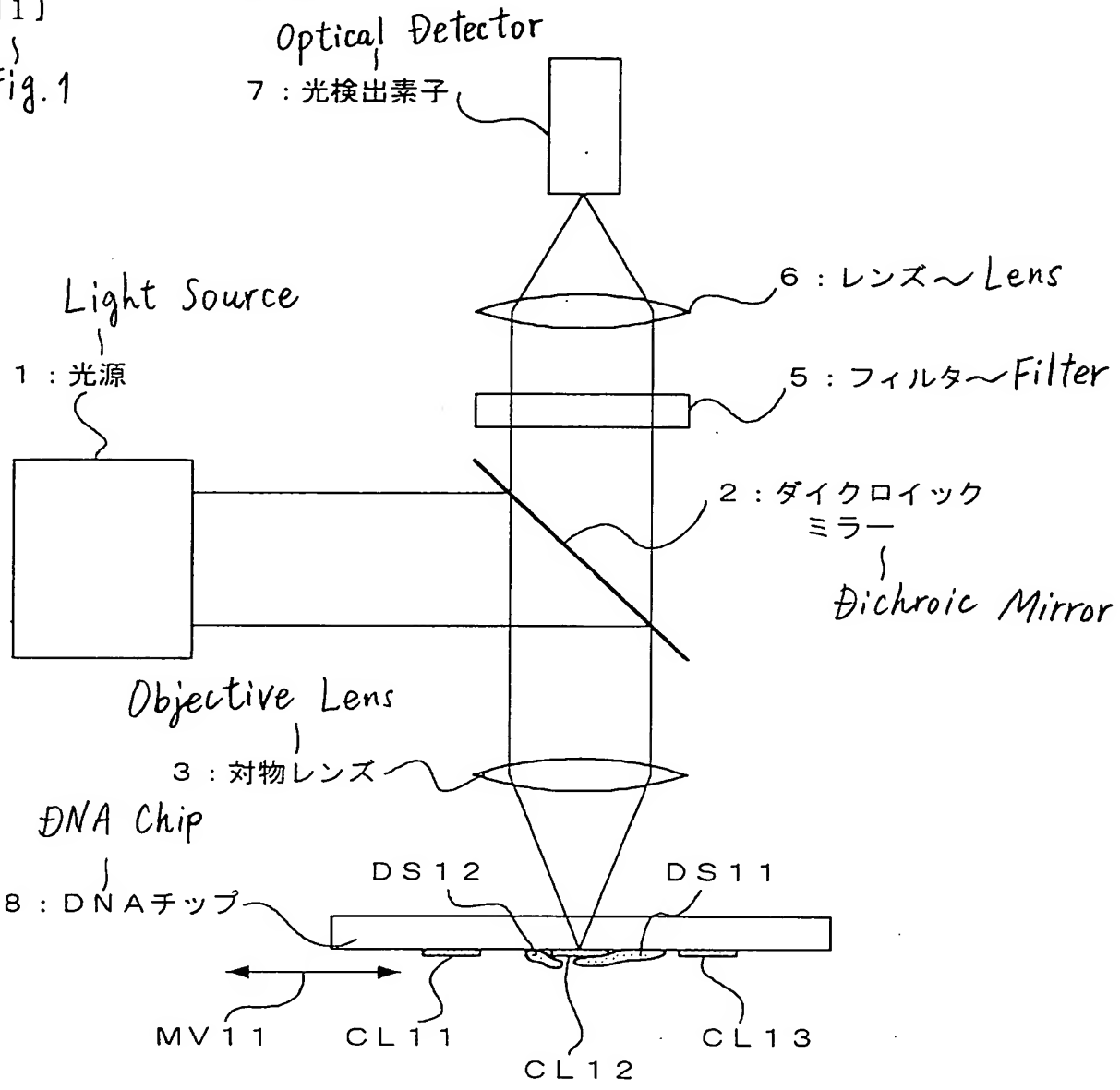
[Explanations of Letters or Numerals]

1	Light source
2	Dichroic mirror
3	Objective lens
4, 8	DNA samples
5	Filter
6	Lens
7	Optical detector
9	Solid immersion lens (SIL)
10	Anti-reflection coating

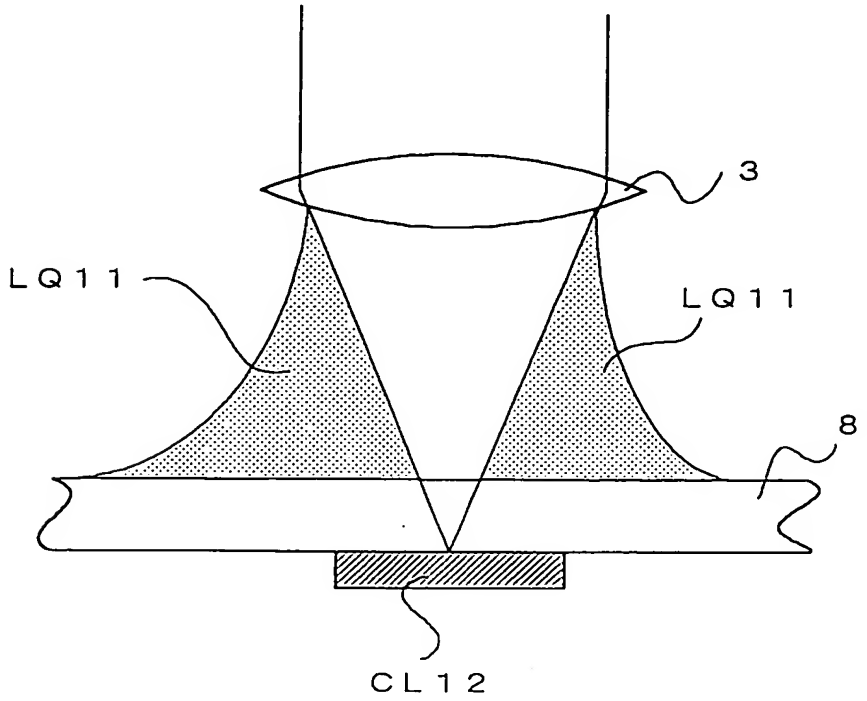
【書類名】～Document Name　図面～Drawings

【図１】

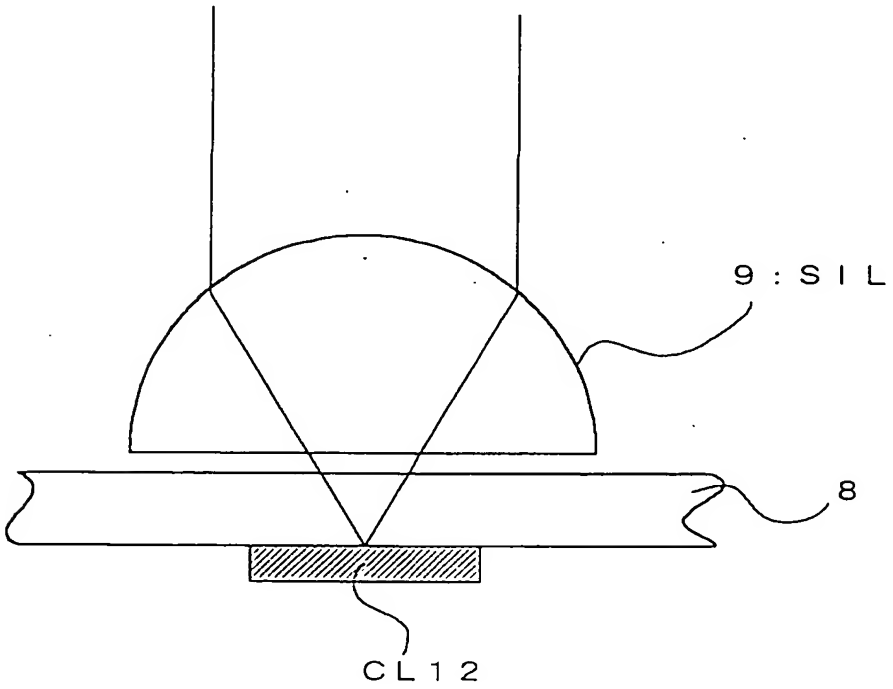
Fig.1



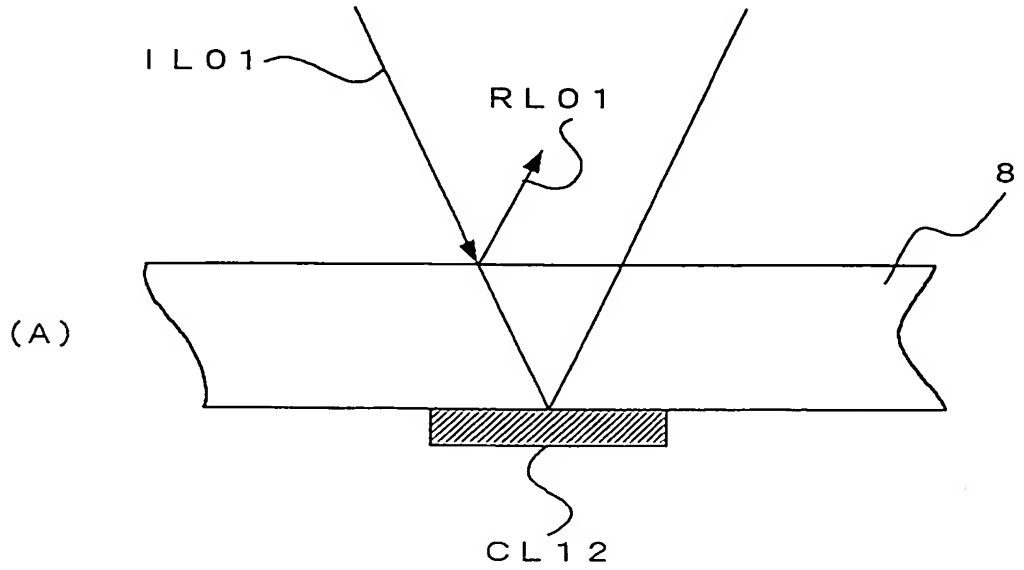
【図２】～Fig.2

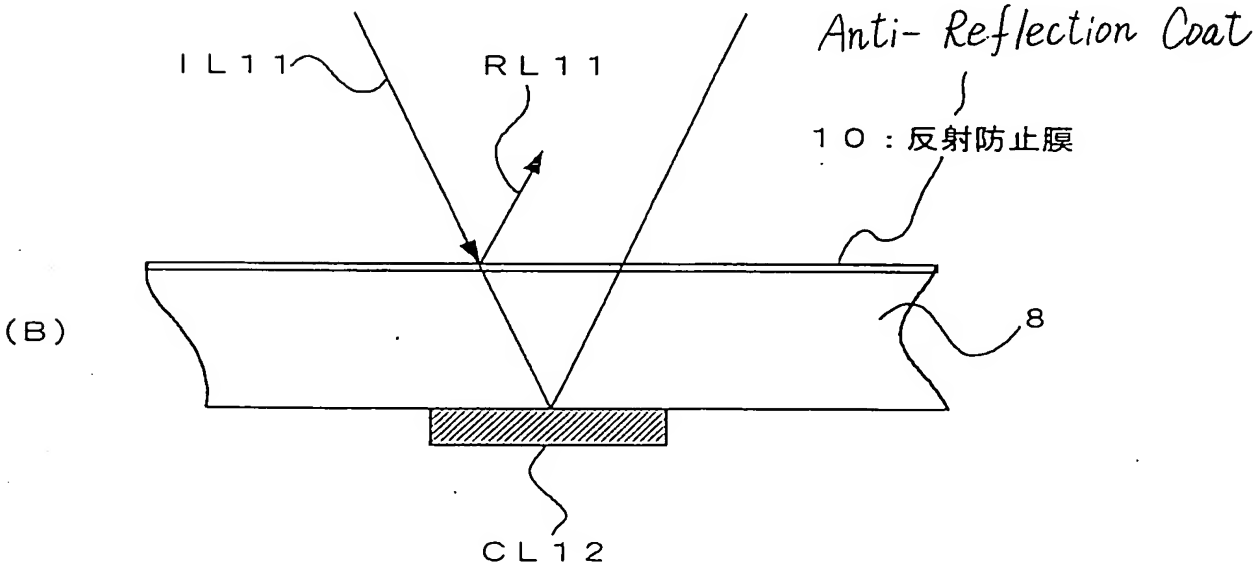


【図３】～Fig.3

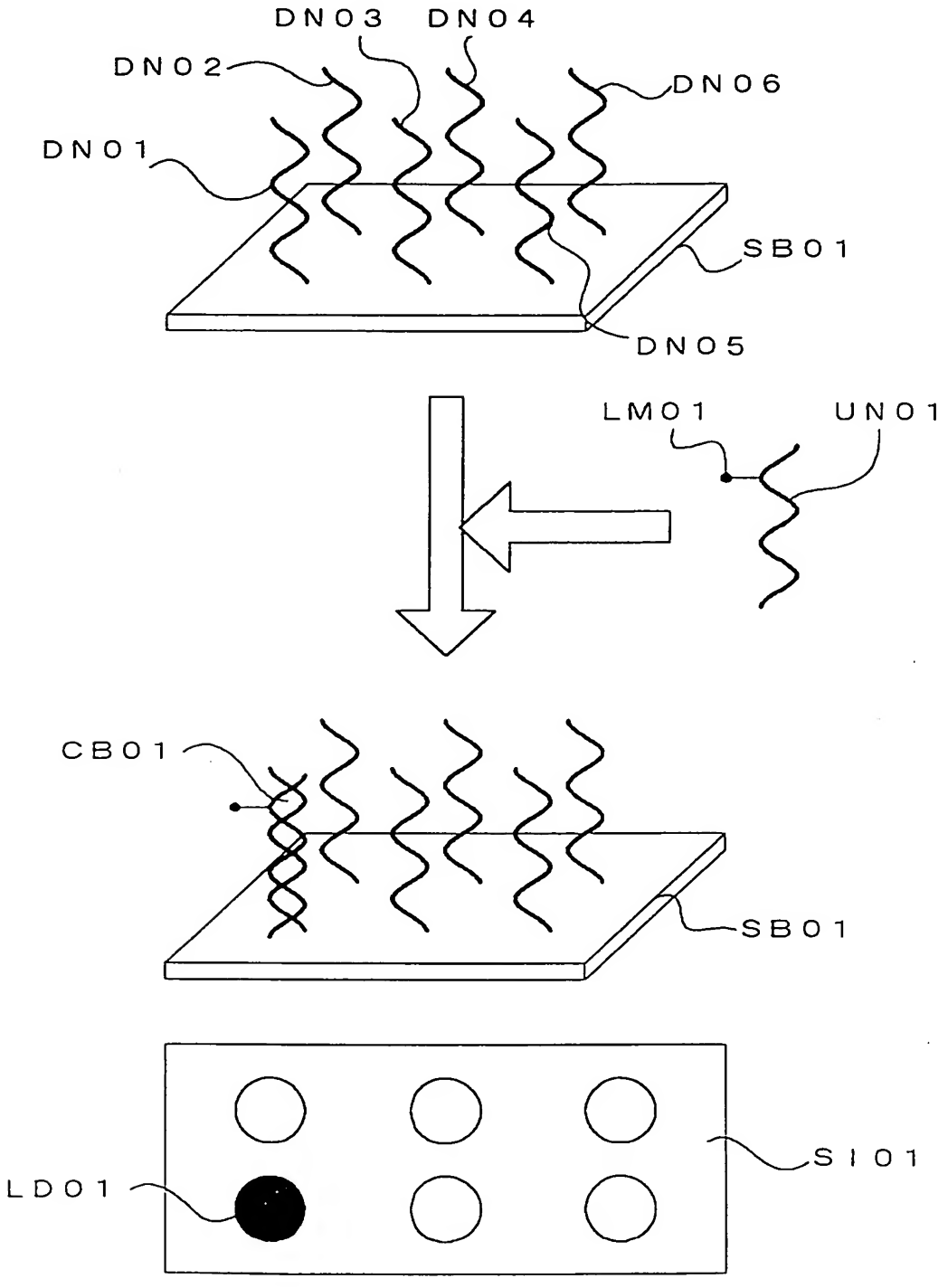


【図４】～Fig.4

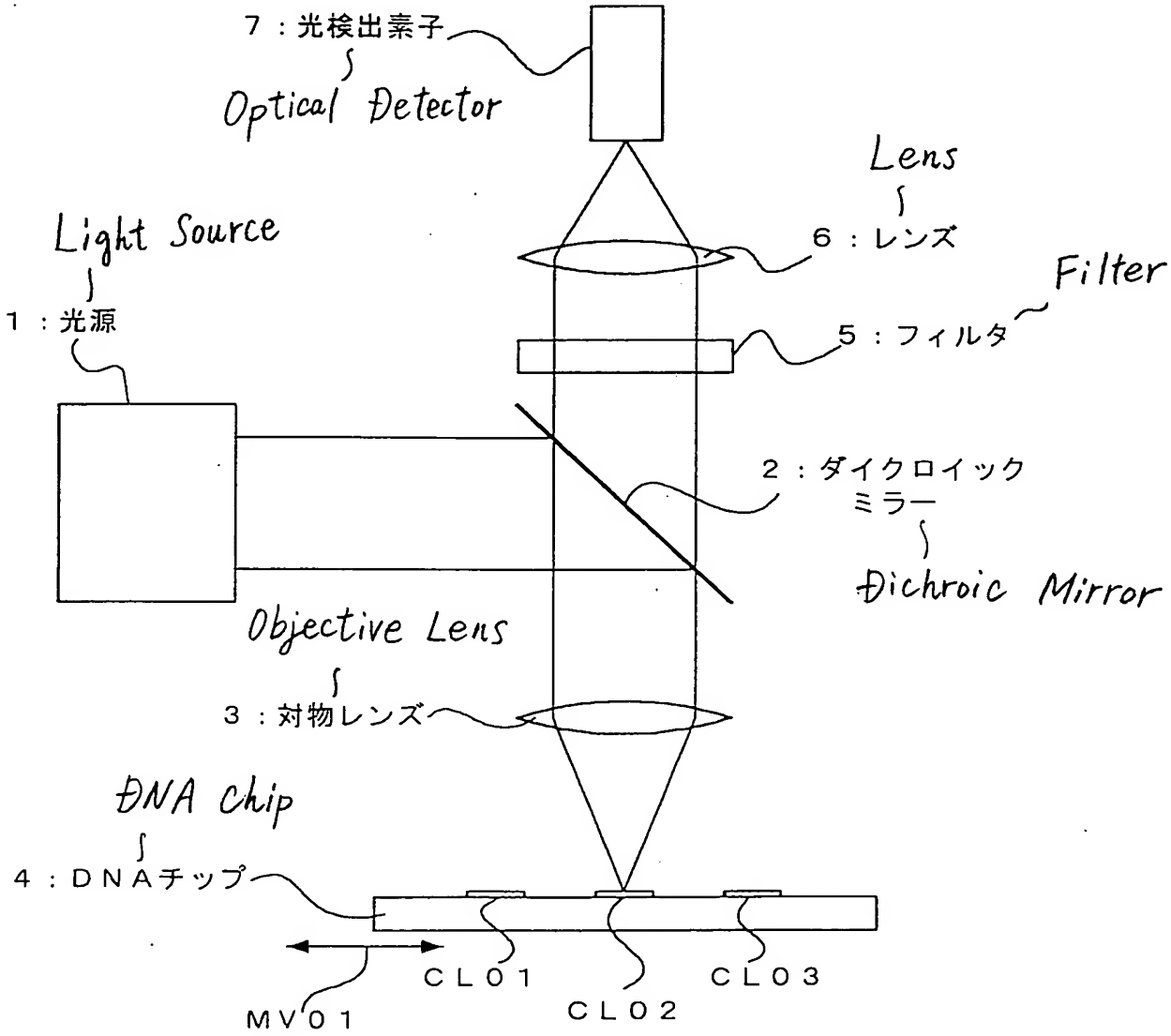

ＩＬ０１
ＲＬ０１
8
(A)
ＣＬ１２


ＩＬ１１
ＲＬ１１
Anti- Reflection Coat
１０：反射防止膜
8
(B)
ＣＬ１２

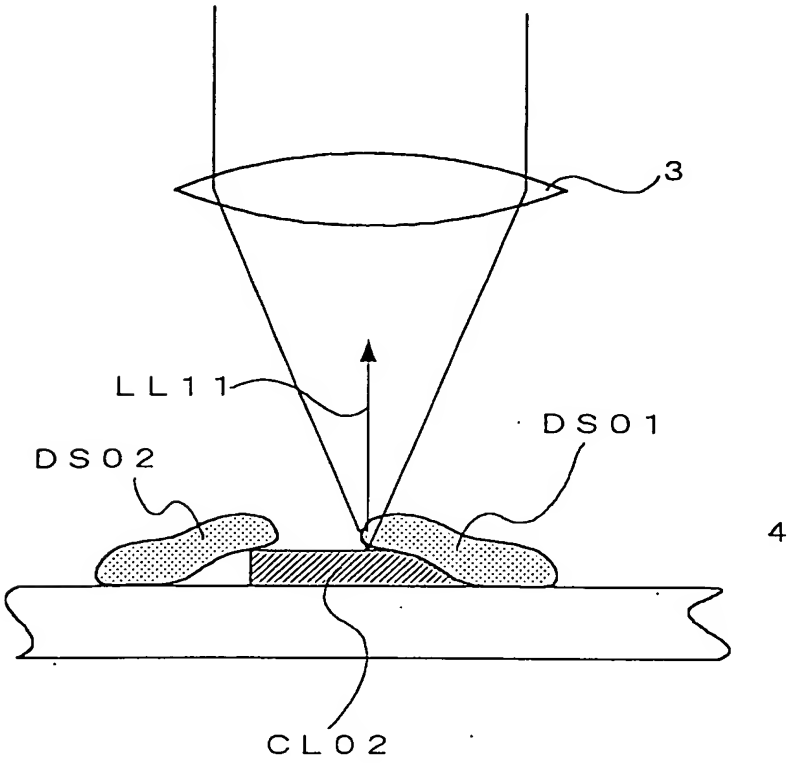
【図5】～Fig.5


DN03
DN04
DN02
DN06
DN01
SB01
DN05
LM01
UN01
CB01
SB01
LD01
SI01

【図６】～Fig. 6


７：光検出素子
Optical Detector
Lens
６：レンズ
Light Source
１：光源
Filter
５：フィルタ
２：ダイクロイック
ミラー
Dichroic Mirror
Objective Lens
３：対物レンズ
DNA Chip
４：ＤＮＡチップ
ＣＬ０１
ＣＬ０２
ＣＬ０３
ＭＶ０１

【図７】～Fig.7



[Document Name] Abstract

[Abstract]

[Object of the Invention]

The object of the present invention is to provide a biochip reader whose S/N ratio is superior and whose cost can be reduced.

[Means for Solving Problems]

A biochip reader comprising:

a light source for emitting excitation light;

a dichroic mirror for reflecting the excitation light or allowing the excitation light to pass through the dichroic mirror;

an objective lens for condensing light that has been reflected by or passed through the dichroic mirror onto a biochip and projecting fluorescent light produced at the biochip onto the dichroic mirror;

an optical detector for detecting the fluorescent light; and

a lens for condensing the fluorescent light that has been reflected by or passed through the dichroic mirror onto the optical detector,

wherein the biochip is configured using a transparent substrate allowing for passage of the excitation light and the fluorescent light and the excitation light is irradiated from one side of the biochip opposite to the side where samples are arranged.

[Chosen Drawing] FIG. 1